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AGRICULTURAL EXPERIMENT STATION
CONDUCTED JOINTLY BY THE
STATE DEPARTMENT OF AGRICULTURE
AND THE
NORTH CAROLINA STATE COLLEGE OF
AGRICULTURE AND ENGINEERING
RALEIGH AND WEST RALEIGH

INHERITANCE OF SEX IN VITIS ROTUNDIFOLIA

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1In coöperation with the U. S. Department of Agriculture, Bureau of Plant Industry.

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4In coöperation with the U. S. Department of Agriculture, Office of Public Roads and Rural Engineering.

FOREWORD

Vitis rotundifolia, the Muscadine grape, has been the subject of investigation by the Department of Horticulture of the North Carolina Agricultural Experiment Station for a number of years, with a view to the determination and the discovery of facts relating to its origin, its characteristics, and the laws which govern their transmission. The results of such research should serve as an addition to our plant-breeding knowledge and help to form a substantial basis for the improvement of this class of grapes.

The results which have been accomplished are set forth in the following bulletins:

1. "Scuppernong and Other Muscadine Grapes: Their Origin and Importance," by F. C. Reimer, Bulletin No. 201, N. C. Agr. Exp. Station. 1909.

2. "Self-Sterility of the Scuppernong and Other Muscadine Grapes," by F. C. Reimer and L. R. Detjen, Bulletin No. 209, N. C. Agr. Exp. Station. 1910.

3. "Breeding Rotundifolia Grapes: A Study of Transmission of Character," by F. C. Reimer and L. R. Detjen, Technical Bulletin No. 10, N. C. Agr. Exp. Station. 1914.

The present bulletin deals with the problems of transmission of sex and constitutes the fourth contribution to this series of reports of work accomplished.

J. P. Pillsbury,
Horticulturist.

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INHERITANCE OF SEX IN VITIS ROTUNDIFOLIA

FIRST REPORT

By L. R. Detjen, Assistant Horticulturist.

I. Introduction

All commercial varieties of rotundifolia grapes of the present day are self-sterile. Because of this fact, staminate vines are required in the vineyards, especially true of large commercial vineyards, in order to produce profitable crops. Just what proportion of the vines should be staminate in order to insure maximum crops is at present an open question. As recommended by Reimer and Detjen (1)* one staminate vine should be planted for every eight fruit-bearing vines. This means that one vine out of every nine in the vineyard will bear no fruit, or, in other words, eleven and one-tenth per cent (11.1%) of the vineyard will be unproductive as far as fruit production is concerned. By planting staminate vines in the vineyard, land which is capable of producing fruit is given over to the production of pollen alone. Indispensable as pollen is, it is not the ultimate object of the fruit grower, and hence that land which is occupied by the staminate vines represents a direct loss.

By replacing each staminate vine with one which bears both pollen and fruit on the same vine, not only would pollen for cross-pollination be produced, but also a crop of fruit. Hence, if every ninth vine in the vineyard could thus be suddenly transformed from an indirect to a direct producer, the proceeds of the land might be increased eleven and one-tenth per cent.

If it were possible to replace all of our fruit-bearing varieties with self-fertile vines, ideal conditions respective to pollination would prevail in the vineyard. Each pistil in the flower would be surrounded by an abundance of pollen, and pollination itself would become independent of insect activities. Hence the crops of fruit would become more regular, irrespective of seasonal and climatic conditions, and the financial returns might be increased correspondingly.

*Reference to cited literature is made by number.

II. MORPHOLOGICAL DIFFERENCES IN THE FLOWERS OF VITIS ROTUNDIFOLIA

Grape flowers can be divided into three general classes or types:

1. The perfect hermaphrodite flower.
2. The staminate or "male" flower.
3. The imperfect hermaphrodite or "female" flower.

FIG. 1.—Three different types of flower clusters and flowers; on the left, staminate; in the middle, hermaphrodite with reflexed stamens; on the right, hermaphrodite with upright stamens. Natural size.

These types may be briefly described as follows:

After the petals, or "caps" as the united petals of a grape flower are called, have fallen, the perfect hermaphrodite flower consists essentially of a normal pistil and five or more upright stamens. Each pistil is functional and under proper conditions is capable of setting and developing a normal fruit. The stamens are strong and healthy. The filaments equal or exceed the combined length of the ovary, style, and stigma. They are stout and fairly symmetrically developed about their longitudinal axes. At the base of the ovary and between the stamens are situated the nectar glands, which secrete nectar, attractive to insects.

The staminate or "male" flowers are hermaphrodite flowers in which the pistils have been almost or wholly suppressed. It is almost superfluous to say that such flowers never produce fruit.

The imperfect hermaphrodite flowers are hermaphrodite flowers which are abnormal in one or more of the floral parts. In this type of flower the pistil has become more strongly developed by an enlargement of the ovary, a corresponding thickening of the style, and an enlargement of

the stigmatic surface. The stamens have undergone a marked transformation in both their form and usefulness. The filaments no longer stand truly upright, as do those in the normal flower, but are more or less recurved or reflexed. According to Dorsey (2), this is due to the unsymmetrical development about their longitudinal axes. In length and in diameter they have suffered material degeneration. Very often their length is considerably less than the greater diameter of the anthers, and their transverse diameter has been so reduced that the filaments bear very much of a resemblance to fine, silken threads. The pollen that is produced by these stamens, instead of being plump and smooth, appears shrunken under the microscope [Reimer and Detjen (1)], and, when seen in quantity, very much resembles a heap of dented and collapsed tennis balls. It is needless to say that such pollen is unviable under the best of conditions, and therefore absolutely worthless. Hedrick and Anthony (3) state that many varieties of grapes with the reflexed type of stamen produce some normal viable pollen grains among the usual misshapen and abortive kind. Such mixed pollen may yet be found with some varieties of rotundifolia grapes; but, to date, many varieties and seedlings of this species have been examined and no normal pollen has ever been found associated with the reflexed type of stamen. The nectar glands that are found in the flowers with the reflexed type of stamen are well developed; they secrete nectar and emit a very fragrant odor which is attractive to many insects.

Such, in general, are the characteristics of the three types of flowers in the genus *Vitis*. Intergrading types are often found, but these will receive special consideration elsewhere.

III. Introduction of Hermaphrodite Vines With Upright Stamens

In the Summer of 1910, while the study of self-sterility of Vitis rotundifolia was in progress, Professor F. C. Reimer (1) discovered a wild vine with upright stamens growing in a plum thicket about one mile from the city of Raleigh, N. C. At the time of the discovery the vine bore a fairly good crop of fruit and, although late in the season, a few flower clusters. It was the flowers on these clusters that commanded attention.

Bagging experiments conducted immediately upon the discovery of the vine gave results indicative of self-sterility. The more extended work of the following two seasons gave similar results.

Although the data obtained in the work of these three seasons pointed very strongly toward self-sterility, a belief in the self-fertility of the vine persisted and evidence secured in later years substantiated the correctness of this supposition.

The strongest evidence that was available in favor of the self-fertility of the vine was the fact that when it was discovered it bore a fairly good crop of fruit. While none of the flower clusters that were bagged in

the two succeeding years set fruit, neither did the flower clusters that grew out in the open produce any berries, although exposed to all chances for cross-pollination. Branches were also bagged and cross-pollinated with pollen from a light-colored staminate vine in order to test out the assumption that with this vine cross-pollination is absolutely necessary, but no fruit developed in any of these bags.

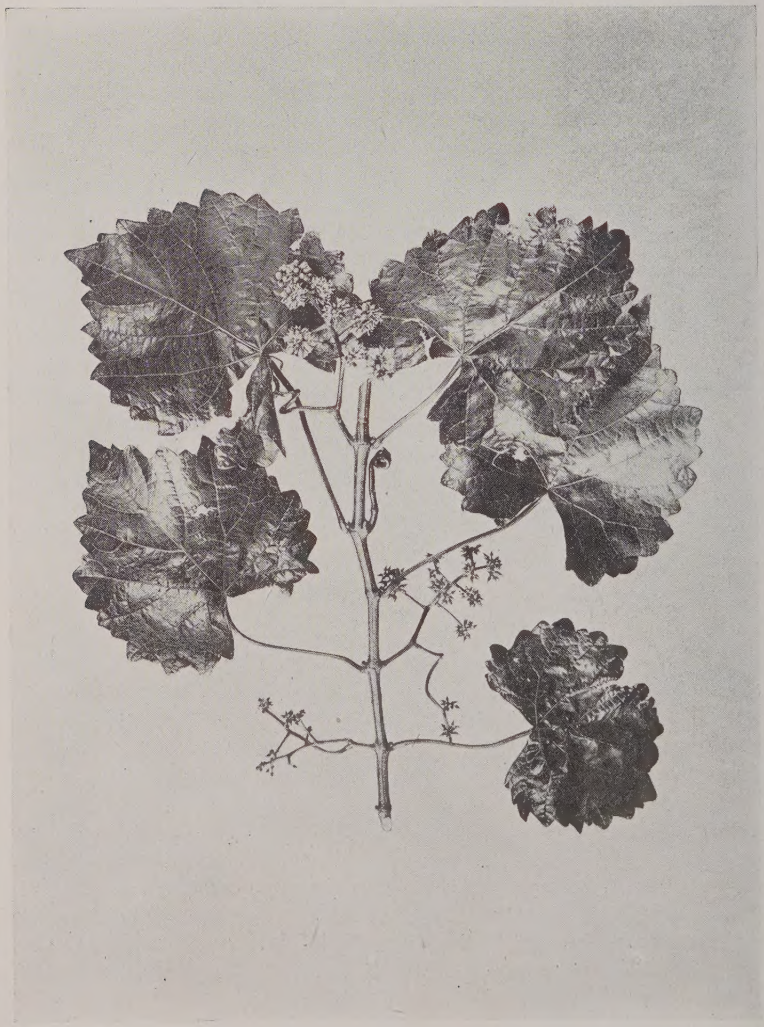

FIG. 2.—Lower section of a flowering branch of Hope. The two lower clusters have shed their stamens recently while the upper cluster is in full bloom. Reduced.

All of this evidence pointed not toward sterility because of defective pollen, but toward partial sterility, probably due to imperfectly developed pistils. The fact, however, that at one time a good crop of fruit had been produced was sufficient evidence that some day, perhaps, the vine might again return to fruitfulness, and this, as we shall see, has since happened.

In the meantime, as this vine was so full of promise, it was called "Hope," in commemoration of the hope and desire to find a self-fertile vine which might become the founder of a new race of rotundifolia grapes.

FIG. 3.—Two clusters of Hope. The cluster on the left is just beyond the blooming stage; note the number and the character of the pistils. The cluster on the right is in full bloom; note the upright stamens and the character of the pistils. Natural size.

Two other self-fertile seedlings, one a light-colored rotundifolia vine, origin not mentioned, and the other a dark-colored vine, a cross between Eden and a staminate vine, are reported by Husmann and Dearing (4).

These three vines are the only original ones mentioned as bearing flowers with upright stamens and with normal viable pollen. To these we look with confidence for the introduction and development of self-fertility among our varieties of rotundifolia grapes.

Description of the Hope vine; Floral Characters.—In general, the vine characters of Hope are typical of such as are found in the rotundifolia species. The plant is only moderately vigorous and is subject to the ravages of Black Rot (*Guignardia bidwellii*) in about the same degree as is the variety, Scuppernong. The main difference between this and other vines of the same species, however, lies in the floral characters. The flowers generally have neither the suppressed pistils of the staminate vines nor the recurved stamens of the common fruit-bearing vines, but they possess a combination of upright stamens with an abundance of healthy viable pollen, similar to that of typical staminate vines, and fairly well developed pistils, which upon fertilization are generally capable of developing into normal fruit. These pistils are not as large as those of other fruit-bearing varieties, neither is the stigma so well developed (Plate I, Fig. 2); still, the pistils are functional, and average sized fruit crops are often produced.

Occasionally, on some poorly nourished branch, which generally may be found on the lower or central portion of the vine, flower clusters are observed with some or all of the flowers representing various stages of development from the totally suppressed pistils of staminate flowers to the fairly well developed pistils of perfect flowers on other parts of the vine.

The filaments of the stamens in all of the flowers of this vine have the same strength, diameter, and upright position as do those in flowers of staminate vines. In short, the flowers of this vine might be termed staminate flowers whose pistils have reached a point in regeneration where they have become functional.

While the Hope vine is not as prolific in flower clusters as are some of our best staminate vines, nevertheless, it is quite representative of the average staminate vine. One or two flower clusters are usually produced on a cane, while three and four, although less often observed, are not at all of rare occurrence. Frequently, also, especially when the vine grows vigorously, many branches are produced which are absolutely barren.

The flower clusters of this vine are of fairly large size, considering the species. In fact, they are larger than those from the average staminate vine, and from external appearances, before the flowers begin to bloom, the clusters are seen to closely resemble those from a fairly good staminate vine. The number of buds on the cluster varies considerably, just as do those from all other vines. Countings made in the field place the number of buds per cluster anywhere between 100 and 300. Our best staminate vine produces clusters with buds ranging in number all the way from 200 to 384. It is probable that none of the fruit-bearing vines with reflxed stamens* produce clusters with more than 120 flowers,

*Eden and San Monta are here excepted, because the former is considered a Munsoniana hybrid and the latter, supposedly a hybrid with a bunch grape, can probably be traced to a similar origin.

since the greatest number of flowers per cluster that has ever been recorded is 113. The vine which bore this cluster is a white fruited rotundifolia seedling found growing wild in the woods near West Raleigh, N. C.

FIG. 4.—A typical fruiting branch from the Hope vine. Note the small fruit clusters. Reduced.

The fruit clusters of Hope are very small; one, two, three, and four berries usually constituting the average cluster. One would think that a cluster bearing from 200 to 300 flowers ought to produce a fairly good-sized fruit cluster, but, contrary to expectation, only a very few berries

FIG. 5.—The largest cluster of fruit that was ever found on the Hope vine. The flowers of this cluster were not protected from cross-pollination. Natural size.

ever set on each of these clusters. The largest bunch of berries that was ever observed on this vine consisted of only ten berries, while the largest bunch of rotundifolia grapes ever observed by the writer was composed of thirty-five berries.* The number of berries per cluster varies with the general care of the vine.

The number of fruit clusters on the vine depends on many factors, among which the actual number of flower clusters produced, the degree of development of the pistils, and the amount of shedding that takes place soon after the flowers are fertilized are important items and demand consideration.

Self-fertility of Hope Vine.—In order to determine the exact status of the Hope vine in regard to the question of self-fertility, the following work was carried out:

In 1910, as soon as the vine was discovered, although this happened late in the season, two flower clusters which were still in bud were observed and immediately bagged. These bags were left undisturbed until all of the buds had bloomed, and later, upon examination of the contents of the bags, no fruit was found to have developed. The other parts of the vine bore an average crop of fruit which was produced from the normal flowers of the season.

In 1911 the experiment was repeated and, because of a greater supply of flower clusters, thirty-five branches were bagged and left to their fate. Not one of these thirty-five branches produced as much as a berry, but neither did the other parts of the vine bear any fruit.

In 1912 ten more branches, with two or more clusters to the branch, were covered. Although the vine bloomed profusely, no fruit was produced that year either in the bags or on the exposed branches of the vine.

In order to determine whether this vine failed to set fruit because of an existing uncongeniality between the pistils and the pollen of the same vine, or because of the lack of viable pollen, nineteen branches were bagged in 1911 and cross-pollinated with fresh pollen from a light-colored staminate vine. This pollen, although from a staminate vine, had no more effect on the setting of berries than did the pollen from the same vine, for no fruit was produced in the bags. The pistils, therefore, seemed to be faulty and incompetent to set fruit at times, regardless of the kind and source of pollen.

In 1913 only two berries were observed on the whole vine, and in 1914 only a handful, less than a dozen berries, constituted the total crop of fruit.

These negative results pointed very strongly toward sterility of the vine. It seems that for some unknown reason the vine appeared to have reverted all at once from a fruit-bearing vine to one that functions essentially as a staminate vine.

*In 1914 a Luola vine produced an exceedingly heavy crop of fruit, and among the many large clusters one was found composed of 35 berries.

In the spring of 1911 a layer of this vine was transferred to the College grounds in order to have the vine under better observation and also to preserve the strain in case destruction should threaten the original vine. This precautionary measure was indeed a fortunate one, because during the spring of 1916 the original vine was destroyed by fire.

FIG. 6.—Two fruit clusters from the Hope vine whose flowers were protected from cross-pollination by having been inclosed in cloth bags. Natural size.

The layer that was transplanted to the vineyard grew well, and in 1912 ten branches were bagged before any buds had opened. Again, none of these flower clusters set fruit, and no fruit developed on the exposed branches of the vine.

In 1913 ten branches with a total of twenty flower clusters were carefully bagged, and this time eight of the ten bags contained some fruit. The whole vine had a fair sprinkling of fruit.

In 1914 twenty branches with not less than a total of forty clusters were bagged, and from each of these twenty bags some ripe fruit was harvested in the following September. The largest fruit cluster that was taken from these bags consisted of eight berries, while the largest cluster that was ever borne by the vine consisted of only ten berries.

In 1915 ten more branches with no less than twenty flower clusters were bagged, and fruit was harvested from eight of these bags in the fall of the year. Again there was a fair crop of fruit on the exposed parts of the vine, and every year since then a fairly good crop of fruit has been produced.

TABLE No. 1

A TEST OF SELF-FERTILITY OF HOPE VINE

Variety	Year	Number Branches Bagged	Number Bags Containing Fruit	Number Bags Containing No Fruit
Original vine, Hope	1910	2	0	2
	1911	35	0	35
	1912	10	0	10
Transplanted vine, Hope	1913	10	8	2
	1914	20	20	0
	1915	10	8	2

From the results of these bagging experiments we learn these important facts:

1. That the pollen of this vine, Hope, can under normal conditions fertilize the flowers on the same vine and on the same flower cluster.

2. That with proper cultivation and care this vine assumes the character of a hermaphrodite vine, while with neglect its pistils gradually cease to function and the vine assumes the general rôle of one that is staminate.

3. That cultivation, pruning, and general care exert a marked effect on the size and the regularity of the crop.

This last point is of considerable economic importance, because the general belief is that varieties of rotundifolia grapes can endure considerable neglect and still produce good crops.

IV. INVESTIGATIONS REGARDING THE INHERITANCE OF TYPES OF FLOWERS IN VITIS ROTUNDIFOLIA

With the discovery of the variety, Hope, we find that Vitis rotundifolia produces three distinct types of flowers instead of two. The important question now arises as to how and in what manner these three types of flowers are transmitted to the progeny of the first generation. Hedrick and Anthony (3), from tabulations of work done with many seedlings of Euvitis, report the following ratios:

Upright stamens × upright stamens=4.3 upright: 1 reflexed
Reflexed stamens × reflexed stamens=1.2 upright: 1 reflexed
Reflexed stamens × upright stamens=1. upright: 1 reflexed
Upright stamens × reflexed stamens=?

Whether these ratios are also representative of the laws that govern the transmission of types of flowers in Vitis rotundifolia is a question deemed worthy of investigation.

The method of procedure naturally resolves itself into a study of the progeny of crosses between vines with different types of flowers. Since there are flowers of three distinct types, namely, staminate flowers with upright stamens, hermaphrodite flowers with upright stamens, and hermaphrodite flowers with reflexed stamens, we may expect to make five different combinations, as follows:

1. Hermaphrodite flowers with reflexed stamens × staminate flowers (with upright stamens).
2. Hermaphrodite flowers with reflexed stamens × hermaphrodite flowers with upright stamens.
3. Hermaphrodite flowers with upright stamens × hermaphrodite flowers with upright stamens.
4. Hermaphrodite flowers with reflexed stamens × hermaphrodite flowers with reflexed stamens.
5. Hermaphrodite flowers with upright stamens × staminate flowers (with upright stamens).

The types of flowers that were observed in the progeny resulting from the first cross (hermaphrodite flowers with reflexed stamens × staminate flowers with upright stamens) has been partially discussed in Tech. Bul. No. 10 of this Station (5). Since that time more extensive and important data has been collected.

The following table shows the results so far obtained:

TABLE No. 2

SEX RATIOS AMONG PROGENY WHEN HERMAPHRODITE FLOWERS WITH REFLEXED STAMENS ARE CROSSED WITH STAMINATE FLOWERS

Female Parent	Male Parent	Number of Vines Staminate	Number of Vines Hermaphrodite With Reflexed Stamens	Number of Vines Hermaphrodite With Upright Stamens
Scuppernong	Light Male, No. 1	11	8	0
Scuppernong	Dark Male, No. 1	329	307	0
Scuppernong	Light Male, No. 2	507	494	0
James	Light Male, No. 2	59	64	0
Thomas	Light Male, No. 2	30	33	0
James	Light Male, No. 2	169	182	0
James	Light Male, No. 1	42	36	0
James	Light Male, No. 2	48	39	0
Flowers	Light Male, No. 2	103	82	0
Thomas	Light Male, No. 2	51	38	0
Scuppernong	Dark Male, No. 3	20	25	0
James	Dark Male, No. 3	9	6	0
James	Dark Male, No. 3	21	18	0
Thomas	Dark Male, No. 3	22	17	0
Latham	Light Male, No. 2	3	2	0
Thomas	Dark Male, No. 3	18	10	0
Thomas	Light Male, No. 2	15	14	0
Memory	Light Male, No. 2	5	6	0
Mish	Light Male, No. 2	2	2	0
Mish	Dark Male, No. 3	1	1	0
James	Light Male, No. 2	1	0	0
James	Light Male, No. 2	0	1	0
Scuppernong	Light Male, No. 2	4	2	0
Scuppernong	Light Male, No. 2	4	2	0
Seedling	Light Male	35	23	0
Totals		1,509	1,420	0

From the above table we learn two things: (1) That whenever hermaphrodite flowers with reflexed stamens are crossed with staminate flowers, only two tpes of flowers are observed in the progeny, namely, staminate flowers and hermaphrodite flowers with reflexed stamens (Plate I, Fig. 1, and Plate 2, Figs. 1-3). Not one seedling out of 2,929, representing the progeny of such a cross, has produced hermaphrodite flowers with upright stamens.* (2) That the ratio of the staminate flowers to the hermaphrodite flowers with the reflexed stamens is as 1509:1420 or 105:1.

The second point under consideration is a study of the progeny resulting from hermaphrodite flowers with reflexed stamens when crossed with hermaphrodite flowers with upright stamens.

In 1911 five branches of a Scuppernong vine were bagged and later cross-pollinated with pollen from Hope as the male parent. In the

*Husmann and Dearing (4) report the finding of two fruit-bearing seedlings with upright stamens among the progeny of vines which bear the reflexed stamens when crossed with staminate vines.

following autumn a normal crop of fruit was gathered from these bags and a total of 108 seeds were harvested.

In 1912 the following similar crosses were made: Scuppernong × Hope, Thomas × Hope, and James × Hope. In each case the Hope vine was used as the staminate parent of the cross, and every cross seemed to be a congenial one, since a normal crop of fruit was harvested from these bags in the fall. A total of 1,351 seeds were obtained from the cross between Scuppernong and Hope, 1,117 seeds from the cross between Thomas and Hope, and 333 seeds from the cross between James and Hope.

In 1913 the variety Flowers was crossed with pollen from the Hope vine, and in all 309 seeds were harvested.

FIG. 7.—A flower cluster with reflexed stamens from a hermaphrodite vine and a flower cluster from a staminate vine. Note the comparative sizes and the characteristic type of each. Natural size.

In 1914 a light-colored seedling of James (F-13) was crossed with Hope and 149 seeds obtained. In the same year a dark seedling of Scuppernong (P-48) was crossed with pollen from Hope and 216 seeds obtained. All of these seeds were planted, and the resulting seedlings were later transplanted to nursery rows for study.

The following table gives the results obtained to date as to the different types of flowers that were observed among the progeny of the aforementioned crosses:

From this table we learn (1) that in every cross where Hope was used as the male parent on vines with reflexed stamens, only hermaphrodite vines have been obtained. Not one staminate vine has as yet appeared in any of these lots of seedlings.

TABLE No. 3.

SEX RATIOS AMONG THE PROGENY OF HERMAPHRODITE VINES WITH REFLEXED STAMENS CROSSED WITH HERMAPHRODITE VINES WITH UPRIGHT STAMENS

Year	Female Parent	Male Parent	Number of Seedlings Surviving	Staminate Flowers	Hermaphrodite Flowers Upright Stamens	Hermaphrodite Flowers Reflexed Stamens	Sex Not Determined
1911	Scuppernong	Hope	57	0	24	33	0
1912	Scuppernong	Hope	661	0	324	328	9
1912	Thomas	Hope	444	0	197	228	19
1912	James	Hope	158	0	79	74	5
1913	Flowers	Hope	104	0	39	53	12
	Totals		1,424	0	663	716	45

FIG. 8.—A typical hermaphrodite flower cluster with upright stamens. F_1 generation seedling of Thomas × Hope (three-year-old vine). Natural size.

By glancing at the record of the first lot, Scuppernong × Hope, the seeds of which were collected in 1911, we learn that of the 57 surviving vines, all are hermaphrodite in character and none are staminate.

In the second lot, where Scuppernong was again crossed with Hope, and seeds gathered in 1912, we see a repetition of the results that were obtained in the first lot, but in this case a much larger number of vines was involved. In the third, fourth, and fifth lots, in which Thomas, James, and Flowers were used to make similar crosses, the final results are practically the same. If we take the total of the numbers that were involved, we find that out of 1,424 vines, 1,379 produced hermaphrodite

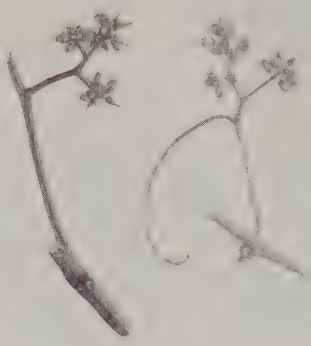

FIG. 9.—Two typical hermaphrodite flower clusters with reflexed stamens. F_1 generation seedling vine of Thomas × Hope (three-year-old vine). Natural size.

flowers, not one produced staminate flowers, and 45 were too weak to produce any flowers at all. These last mentioned vines unquestionably will also bear hermaphrodite flowers when mature.

We also learn (2) that these seedling hermaphrodite vines are of two kinds (Plate 1, Figs. 3-6), those which produce flowers with upright stamens and those which produce flowers with reflexed stamens. The ratio between these two floral types is quite uniform, and appears to be about equally divided. By adding the total numbers, we find that the upright stamen type is to the reflexed stamen type as 663:716, or a ratio of 1:1.07.

Fig. 10.—A typical flower cluster from the variety Flowers. Natural size.

Before drawing conclusions relative to the ratios existing between the different types of flowers, the utmost care should be exercised in getting as complete data as is possible, because very often hermaphrodite vines with the reflexed type of stamens are observed to require one or more years to reach full maturity than staminate, or hermaphrodite vines with upright stamens. This point is well illustrated in the following table.

TABLE No. 4

MISLEADING SEX RATIOS

Parentage	Age of Vines	Total Number Vines	Number Vines Blooming	Staminate Vines	Hermaphrodite Vines—Reflexed Stamens	Hermaphrodite Vines—Upright Stamens
Scuppernong × L. Male	2 years	51	18	16	2	0
Thomas × L. Male	2 years	72	31	23	8	0
James × L. Male	2 years	64	47	27	20	0
James × L. Male	2 years	63	31	17	14	0
Scuppernong × L. Male	2 years	128	66	55	11	0
Scuppernong × D. Male	2 years	644	198	143	55	0
Scuppernong × L. Male	2 years	832	176	157	19	0
Lab. Aest. × Lab. Male	2 years	65	19	19	0	0
Totals		1,919	586	457	129	0
Scuppernong × Hope	3 years	57	8	0	2	6
Scuppernong × Hope	2 years	661	79	0	18	61
Thomas × Hope	2 years	443	108	0	27	81
James × Hope	2 years	157	31	0	9	22
Total		1,318	226	0	56	170

From the first part of this table it becomes evident that the staminate vines in every case outnumber the hermaphrodite vines which bear the reflexed type of stamen, and that the ratio of the total number of such vines at the age of two years from seed is 457 staminate to 129 hermaphrodite vines with reflexed stamens, or 3.5 : 1. But there are still 1,333 vines which have not yet bloomed. It is necessary, therefore, that the types of flowers of all of these be considered before final conclusions are drawn.

The second part of the table shows the numbers of vines of the different types that bloomed at the age of two and three years when Hope was used as a male parent. Again, those plants that bear the upright stamens are in the majority, 170 vines with upright stamens as compared with 56 vines of the type that bears the reflexed stamens, or a ratio of 3 : 1.

Referring to Tables Nos. 2 and 3, we observe that when these same vines are three or more years of age the ratios of the vines that bear upright stamens to those that bear the reflexed stamens have been almost equaled. The ratios in both of these tables no longer stand as 3 : 1, but practically as 1 : 1, the theoretical ratio.

The third point under consideration deals with the ratio of the types of flowers that appear among seedling vines when both of the parent vines produce hermaphrodite flowers with upright stamens.

While testing the Hope vine for self-fertility, a certain number of seeds were secured from fruits that had developed in the cloth bags without the introduction of any foreign pollen. In 1913 sixty-eight seeds were thus secured and planted. From these 68 seeds only 7 plants

were obtained, of which, at the present writing, only 6 are alive. In 1915 two of these vines bloomed and each produced only staminate flowers.

In 1915, 47 seedlings were obtained from the same mother vine, and in 1916, 530 more seedlings from a similar origin but from different vines, were set in the nursery row for observation and study.

In the fall of 1913 a large number of seeds were taken from fruits of the Hope vine which had developed under natural conditions, that is, with the flowers uncovered and subject to any cross-pollination that might take place. From these seeds 142 plants were obtained and some of these have bloomed this year (1916). The types and numbers of vines that have bloomed and been observed are as follows: staminate vines, 21; hermaphrodite vines with upright stamens, 31; hermaphrodite vines with reflexed stamens, 4. The number of vines not blooming was 86. Of these 86 non-blooming vines, it was confidently expected by the writer that 17 vines, by virtue of their size and age, would produce hermaphrodite flowers with reflexed stamens. The remaining 59 seedlings were too small and weak to produce any flowers. If we accept the numbers of these vines as stated, the ratios of the different types is 21:31:21. What the final ratio will be when all of the vines shall have bloomed becomes, at most, an interesting matter for speculation.

TABLE No. 5

THE EXTENT OF NATURAL CROSSING THAT MAY TAKE PLACE AMONG THE SELF-FERTILE SEEDLING VINES OF THE HOPE VINE PROGENY WHEN GROWING IN MASS

Lot	Color Mother Vines	Total Number Seedlings Grown	Number Seedlings Light Color	Number Seedlings Dark Color
1	Light	112	110	2
2	Light	64	58	6
3	Light	59	55	4
4	Light	16	13	3
5	Light	105	88	17
6	Light	20	19	1
7	Light	104	104	0
8	Light	40	38	2
9	Light	96	90	6
Totals		616	575	41

In this work, seeds from light-colored vines only were selected, because, as the factor for coloration is dominant to its absence, the appearance of dark-colored seedlings in any of these several lots would be strong evidence in favor of cross-pollination.

From the evidence presented in Table No. 5, we learn that in eight out of nine lots of seedlings some dark-colored plants have appeared. By way of explanation, it should be said that the dark-colored vines, 547 in number, standing adjacent to the light-colored ones from which the

seeds were taken, were heterozygous for the light and the dark colors. If, then, any crossing took place between the light and the dark vines, the resultant dark-colored seedlings, according to the Mendelian laws, represent only about one-half of the actual number of such a cross. This very meager evidence gives us some idea of the actual amount of intercrossing that takes place among our strain of hermaphrodite vines that have the upright stamen. The ratios of colors that were obtained among these seedlings, however, are not to be taken as an index of the extent of cross-pollination that usually takes place among such seedling vines, but simply as an indication that some crossing has resulted.

The fourth point under consideration deals with the ratios between types of flowers among seedling vines when both of the parents bear reflexed stamens. Since Bul. No. 209 of this Station (1) discusses at some length the self-sterility and the inter-sterility of all rotundifolia grape vines which bear reflexed type of stamen, we understand immediately why such seedling vines have not been produced and, hence, why we cannot establish a ratio between types of flowers resulting from such a cross.

The fifth point under consideration deals with the ratios between types of flowers in the progeny when hermaphrodite flowers with upright stamens are crossed with staminate flowers. In 1915 twenty flower clusters of this Hope vine were carefully emasculated and bagged and later, after about twenty-four hours, viable pollen from a staminate vine was brushed over the pistils. No fruits developed in any of these bags.

The same spring 53 clusters on one of its self-fertile seedling vines were carefully emasculated and, when the pistils became receptive, were pollinated with fresh, viable pollen from a staminate vine. Not one of these flower clusters, however, has produced so much as a berry. At the same time, two large flower clusters on this same self-fertile vine, the flowers of which were not emasculated, were dusted with viable pollen from a staminate vine which is known to be homozygous for dark color. Eighteen berries ripened and a total of 61 seeds were obtained. These seeds were planted in 1916 and from them 59 seedlings were grown. Each of these 59 seedlings were light in color and not one was dark. This indicates that the seedlings were not the result of a cross between two separate vines, but the product from self-fertilized flowers on the mother vine, since dark color of vine is dominant over light color. [Reimer and Detjen (5), Hedrick and Anthony (3)].

In 1916 flowers of twelve more clusters from light-colored self-fertile seedlings were carefully emasculated and later hand pollinated with viable pollen from another staminate vine which is also known to be homozygous for dark color. Not one of these flower clusters produced a berry. At the same time the flowers on 50 or more clusters on similar light-colored, self-fertile seedling vines (all seedlings of Scuppernong and Hope) were emasculated, although less carefully in regard to the selfing of the flowers, and immediately cross-pollinated with pollen from a staminate vine which was known to be homozygous for dark color.

Of these 50 bags, 38 were later removed because they contained no fruits. Twelve bags contained one or more fruits, and from these 142 seeds were secured. These seeds will be duly planted, and the colors of the resulting seedlings were well watched with keen interest.*

V. Correlation Between Type of Stamen and Character of Pollen and Between Size of Flower Cluster and Character of Pollen.

The theory that the reflexed type of stamen in Vitis is usually associated with degenerate, impotent pollen, and with complete or nearly complete self-sterility, is generally conceded by most of the present-day investigators. In like manner the corollary of this theory, that upright stamens are generally an index of normal viable pollen and self-fertility, is also recognized [Dorsey (2), Hedrick and Anthony (3), and Beach (6)].

While this correlation between type of stamen and character of pollen holds quite true (Plates III and IV), the numerical relations that exist between viable and defective pollen grains in the same flower seem to be more or less variable.

Booth (7) says, in effect, that among garden varieties of grapes one can find, with the aid of a microscope, a whole series that produce more or less defective pollen. The percentage of this defective pollen in such varieties ranges all the way from almost none in the self-fertile varieties to 100 per cent in those that are self-sterile. What relation the amount of this defective pollen bears to the type of stamen by which it was produced, he does not make clear.

Hedrick and Anthony (3) have successfully crossed cultivated or garden varieties of *Euvitis* where both of the parents bore the reflexed type of stamen. The same authors have grown numbers of seedlings from such varieties when close pollinated, all of which goes to show that some normal pollen grains are frequently and habitually produced by some varieties that have the reflexed type of stamen.

While such varieties of grapes produce some viable pollen grains, not enough of these are formed, however, to class such vines among the perfectly self-fertile varieties. Barring the effects of ravaging diseases and other external factors, the looseness of a fruit cluster frequently is an indicator of the production of an insufficient amount of viable pollen for perfect fertilization. Most of the varieties with the reflexed type of stamen, however, are completely self-sterile, a fact which is due to their defective and degenerate pollen. This is probably more true of wild vines of the native species than of most garden varieties which are representative of hybridization and selection. Booth (7) says: "In

*Since the manuscript of this bulletin was prepared these 142 seeds have been planted and 39 seedlings have appeared above ground. Of these, 25 are dark in color and 14 are green. The dark seedlings, therefore, are the result of a cross between the light-colored and perfect flowered vine with upright stamens and a dark-colored staminate vine. Later on, these dark seedlings will be transplanted to the nursery row, and when grown the types of their flowers will be duly recorded.

the wild, the grape is dioecious, and fertile pollen borne by the pistillate flower with reflexed stamens is rare, if it occurs at all."

Among the many vines of Vitis rotundifolia, both the cultivated and the wild, that have been studied at this Station, not one case has been discovered where normal fruit is definitely known to have developed from pollen that was produced by the reflexed type of stamen.*

The production of seedless berries must naturally be omitted from the present discussion, because it is not the result of a sexual union between two normal gametes, and it is not the purpose of this bulletin to discuss parthenocarpy in relation to the pollination of flowers.

That all vines of Vitis rotundifolia which bear the reflexed type of stamen are completely self-sterile is borne out by the published results in Bul. No. 209 (1). To these results we can add from Tech. Bul. No. 10 (5), that when 693 seedlings were raised from Scuppernong crossed by a male vine homozygous for dark color, not one light-colored vine appeared in the whole lot. It will be remembered that in crossing varieties of rotundifolia grapes which have the reflexed type of stamen, the usual emasculation process is habitually omitted, because if any fertile pollen were produced, its presence could quickly be detected in the progeny of such a cross by the presence of light-colored seedlings. Likewise, the 62 seedlings from James crossed with the same dark male vine were all dark in color, notwithstanding the heterozygous nature of the James variety with respect to light and dark colors.

The correlation of upright stamens with viable pollen and self-fertility of the grape is also generally recognized. While the introduction of rotundifolia grape vines that bear upright stamens is of recent date [Reimer and Detjen (1), and Husmann and Dearing (4)], all have proved to be self-fertile, at least in varying degrees, and consistent with the small fruit clusters of this type of grape. Hope, our original wild vine, and its vegetative offspring, in some years under favorable conditions, has proven to be self-fertile, as recorded elsewhere in this bulletin.

All of the perfect flowered seedlings of this same vine, the founder of our race of self-fertile rotundifolia grape vines, also produce normal viable pollen like their parent and are self-fertile in the same degree.

Husmann and Dearing (4) report that both of their original seedlings with upright stamens are fruitful and self-fertile. Also, that all of their more recent seedlings of similar character are likewise self-fertile.

In 1915 flower clusters on a number of perfect flowered seedling vines were bagged for a test of their self-fertility. The bags happened to be distributed as follows: one bag on each of 64 different vines, six bags on one special vine, and eighteen on another special vine. The results obtained are presented in the following table.

*The two berries that are mentioned in Bul. 209 of this Station as having developed under closed cloth bags on a Scuppernong vine are regarded, in the light of more recent investigation, as the result of cross-pollination, which must have taken place previously to the bagging operation. That a single flower in the bag which covered several hundred similar flowers should receive sufficient pollen to produce three or four seeds, as was found in this case, while other flowers under the same bag by laws of chance should receive absolutely none, appears almost incredible. It is much easier to conceive of two fertilized blooms having been overlooked in the examination and preparation of the many thousands of buds that were covered in the experiment.

TABLE No. 6

SELF-FERTILITY OF HERMAPHRODITE SEEDLING VINES.

Parentage of Seedling Vine	Number Vines Represented	Number Bags Used	Character Stamens	Number Bags Containing Fruit	Number Bags Without Fruit
James × Hope	20	20	Upright	11	9
Scuppernong × Hope	24	24	Upright	14	10
Thomas × Hope	20	20	Upright	9	11
Totals	64	64	Upright	34	30
Scuppernong × Hope	1	18	Upright	16	2
Thomas × Hope	1	6	Upright	5	1

In 1916 eight special vines were tested for self-fertility by placing ten bags at random on unopened flower clusters of each. The results that were obtained in this test are set forth in the following table:

TABLE No. 7.

SELF-FERTILITY OF HERMAPHRODITE SEEDLING VINES

Parentage	Number Branches Bagged	Character Stamens	Number Bags Containing Fruit	Number Bags Without Fruit
Flowers × Hope	10	Upright	0	10
Flowers × Hope	10	Upright	6	4
Flowers × Hope	10	Upright	3	7
Flowers × Hope	10	Upright	8	2
Thomas × Hope	10	Upright	0	10
Thomas × Hope	10	Upright	6	4
Thomas × Hope	10	Upright	0	10
Scuppernong × Hope	10	Upright	5	5
Total	80	Upright	28	52

In the two preceding tables we observe at a glance that in many cases fruit was produced and obtained without the aid of any cross-pollination. No such results have ever before been observed or obtained when flowers with reflexed stamens were isolated from chances of cross-pollination.

It will be observed, however, that not all of the vines which were included in this test produced fruits in the bags. This, however, is not a strange phenomenon when we consider the fact that each one of the sixty-four vines was represented by the flowers of only one or two clusters contained in only one bag. Even the eight vines on which ten and more clusters were bagged gave evidence that at times no fruits may set in some of the bags, and the vines still show signs of self-fertility by reason of the fruits that developed in the others. The failure of one or all clusters to produce fruit cannot well be regarded as proof of absolute sterility. Any one at all familiar with the rotundifolia species knows how common it is for young, vigorous vines to shed a great many of the

flower clusters without setting fruit when heavily pruned and crowded together as are these vines.

The point is well illustrated in the last table, where all of the eight vines shed some of their flower clusters, and three out of the eight produced no fruits at all inside of the bags. As the season of 1915 was an exceptionally unfavorable one for fruit production in the experimental vineyard, these negative results must be looked upon as indicative only, but a further trial or investigation doubtless will prove these same vines to be self-fertile like the others. All of the vines that set no fruit in the bags were subsequently examined as to the character of their pollen, and in no case did this prove to be anything but normal.

From these results we conclude that upright stamens in rotundifolia grape flowers are always associated with viable pollen, and, if the pistils are normally developed, with a certain degree of self-fertility of the vine.

It might be of interest to know that along with the character of the pollen, and the type of stamens, there is correlated a third character, namely, that of size of flower cluster. The staminate vines produce comparatively large flower clusters; the hermaphrodite vines that bear upright stamens produce flower clusters of a similar size, while the hermaphrodite vines with the reflexed stamens generally produce small flower clusters.

Notwithstanding the fact that the flower clusters of the three different types of vines vary considerably among themselves, it is always possible to determine with comparative ease and accuracy what the type of stamen and the character of pollen will be while the vine is still in bud.

When hermaphrodite vines that bear the reflexed type of stamen are crossed with staminate vines, the resulting progeny can be separated and grouped according to their sexes by means of the size of their respective flower clusters. In like manner the progeny of hermaphrodite vines that bear the reflexed type of stamen, when crossed with pollen from hermaphrodite vines that bear the upright stamens, can be separated and grouped according to the character of their pollen by nothing more than their characteristic flower clusters.

These correlated characters are of considerable importance to the rotundifolia grape breeder, because by means of them a comparatively early selection and segregation of vines can be made with respect to normal viable pollen and self-fertility.

VI. Similarity Between Flower Clusters That Are Borne on Hermaphrodite Vines With Upright Stamens and Those That Are Borne on Staminate Vines.

As the staminate vines when in bud, and before any flowers have opened, can be readily distinguished from the imperfect hermaphrodite vines by the difference in the size and shape of their respective flower clusters (see Fig. 7), so in like manner the perfect hermaphrodite vines

FIG. 11.—Two flower clusters from hermaphrodite vines of the F_1 generation of Flowers × Hope. The cluster on the right bears the upright type of stamen, while the cluster on the left bears the reflexed type of stamen. These two clusters are the largest of their kind that could be found among 103 vines. Natural size.

can be distinguished from the imperfect hermaphrodite vines (see Figs. 8, 9, 10, and 11). But a staminate vine and a perfect hermaphrodite vine when in bud cannot be so easily distinguished from one another, if at all.

A flower cluster on a perfect hermaphrodite vine, like one on a staminate vine, normally consists of a great many flowers crowded together on a more or less lengthened rachis (see Figs. 7, 8, 11, and 12). The largest number of buds that were ever counted in a single flower cluster on a staminate vine was 384 and on a perfect hermaphrodite vine (Hope) 299. This staminate vine, however, is one of the best ever observed by the writer among wild vines, while Hope is the first of its kind that was ever recorded among rotundifolia grape vines.

FIG. 12.—Variations in size of the flower clusters from hermaphrodite vines that bear the upright type of stamen. Natural size.

When perfect hermaphrodite vines become as plentiful as the staminate vines, then by virtue of a wider choice we can expect to find flower clusters on such vines equally as large as those on any staminate vine. Selected perfect hermaphrodite seedlings now in our possession doubtless will produce flower clusters equally as large as those that were observed on the staminate vine, but the average flower clusters on the average perfect hermaphrodite vines are about the same in size as those on similar staminate vines.

VII. The Probable Origin of the Hope Vine

By comparing descriptions of Hope with that of any staminate vine, one can readily see that the two have many things in common. Both vines produce pollen which, when mixed, cannot, by aid of the microscope alone, be distinguished as being the product from two different vines (Plate III, Figs. 1 and 3, and Plate IV, Figs. 1, 2, and 3); the pollen of both is viable and will germinate in sugar solutions; both produce upright stamens of identical size, shape, and appearance (see Figs. 3 and 7); and both produce large flower clusters typical of the staminate vines. The only point of difference lies in the fact that the Hope flowers possess functioning pistils which are not found in any staminate vine.

The seedlings of any hermaphrodite variety that has the reflexed type of stamen, when crossed with Hope as the male parent, are always hermaphrodite vines, but of two kinds: (1) those that bear the upright stamens, and (2) those that bear the reflexed stamens (see Figs. 8 and 9). These types occur in the expected ratio of 1:1, just as those whose male parent is a normal staminate vine. These perfect flowered seedling vines with upright stamens also come to maturity earlier than do those with the reflexed stamens, just as do normal staminate seedlings (see Table No. 4). The whole appearance and behavior of the Hope vine, and of its perfect flowered seedlings, with the exception of the regenerated and functioning pistils (Plate I, Figs. 2, 4, and 6), is that of a normal staminate vine.

Again, some of the flowers on the Hope vine and some on its perfect flowered progeny have been observed to be more or less intermediate between perfect hermaphrodite flowers and staminate flowers (Plate II, Figs. 4-9). Some of the pistils were dwarfed, some rudimentary, and some few were suppressed entirely (see Fig. 14), as is the case of staminate flowers. Such flowers were found to occur more abundantly on some vines than on others, but usually in all cases they were found on the lower and more poorly nourished branches of the vine.

All of this evidence points strongly toward the assumption that this Hope vine was primarily a staminate vine, with a staminate constitution, but one which from some unknown cause has undergone a partial regeneration of its pistils, some of which are able to function.

When flowers on many staminate vines are examined, it will be observed that there exists much variation in the size of the suppressed

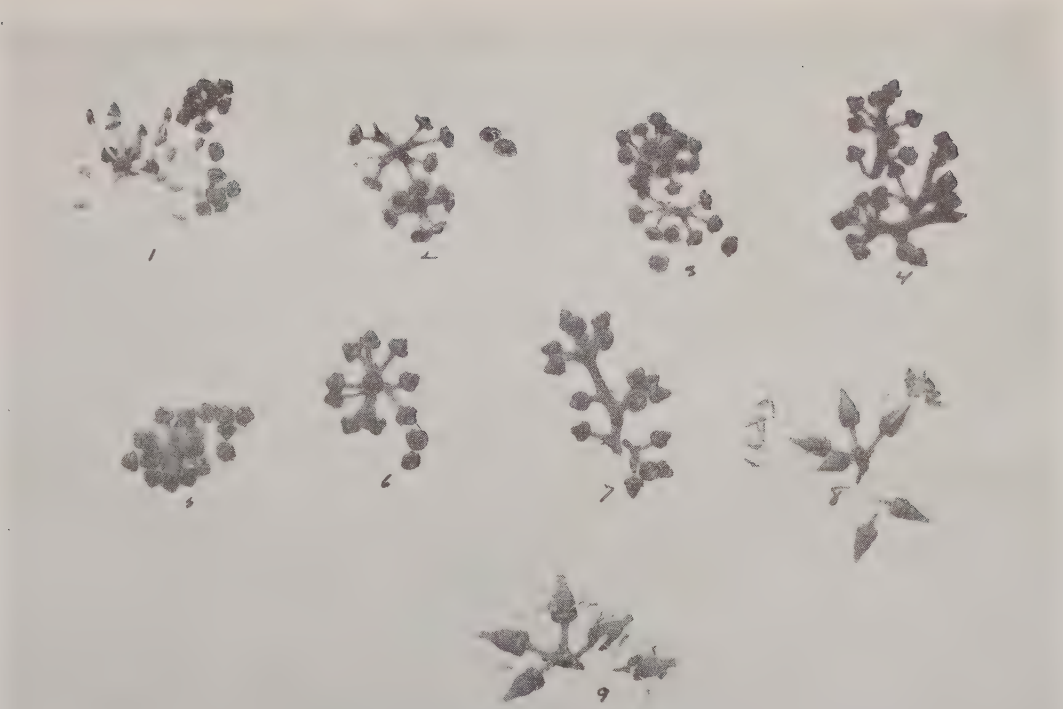

FIG. 13.- Variation in the pistils of Vitis rotundifolia. Numbers 1 to 7, inclusive, represent the variations in the pistillate rudiments which were observed on different staminate vines. No. 8 shows the type of pistil that is found on hermaphrodite vines that bear the upright stamen. No. 9 shows the type of pistil that is found on hermaphrodite vines that bear the reflexed stamen. Natural size.

pistils (see Fig. 13, and Plate II, Figs. 1-3). In fact, many pistillate rudiments in flowers on staminate vines are larger than many similar rudiments in flowers on hermaphrodite vines. With this variation of the pistils in mind, it is not difficult to imagine that by a sudden release from an inhibiting factor these pistils, so long suppressed, have suddenly grown out and become functional.

FIG. 14.—Variations in size of the pistils frequently found on the hermaphrodite seedlings of Hope which bear the upright stamens. Cluster on the left represents the normal type of pistil, while cluster on the right shows considerable reduction in size of the pistils. Natural size.

The strongest link in the chain of evidence yet secured, which points out that the Hope vine is essentially a staminate vine with its suppressed pistils regenerated, lies in the evidence already submitted, to the effect that although fertile when treated with pollen from the same or similar flowers, these pistils are often unfertile when treated with pollen from a staminate vine.

Whether our race of self-fertile hermaphrodite vines represents a progressive step in the line of the evolution of rotundifolia grape flowers is a question that is not so easily proven. The fact that staminate vines show much variation in the size of the suppressed pistils, and that all of the hermaphrodite vines which function as pistillate vines bear stamens and pollen, although functionless, seems to indicate that early in the history of Vitis all of the vines bore hermaphrodite flowers whose stamens and pistils were perfect and functional. Considering the facts of the case in the light of our recent investigations, we may assume our original vine Hope to be a partial reversion to its ancient hermaphrodite ancestors.

VIII. SUMMARY

The investigations regarding the inheritance of self-fertility in Vitis rotundifolia which are being carried on at this Station have led to the following conclusions:

1. That Hope, the first discovered hermaphrodite grape vine of the species Vitis rotundifolia, which bears upright stamens, is self-fertile.

2. That the self-fertility of the Hope vine is variable, and seems to depend on its inner constitution.

3. That floral types in Vitis rotundifolia are transmitted to the progeny in definite ratios. Substituting S for staminate flowers, R for hermaphrodite flowers with reflexed stamens, and U for hermaphrodite flowers with upright stamens, we find that

$$R \times S = 1\,R : 1.06\,S$$
$$R \times U = 1.07\,R : 1\,U$$
$$U \times U = S : R \text{ (ratios not determined)}$$

4. That the Hope vine, and some of its self-fertile progeny, apparently will not cross with staminate vines.

5. That in Vitis rotundifolia the upright stamen in hermaphrodite flowers is correlated with normal, viable pollen, and self-fertility; that the reflexed stamen in hermaphrodite flowers is always associated with defective pollen and self-sterility.

6. That the Hope vine and all of its seedlings, which bear upright stamens, appear and behave essentially like staminate vines except for the facts that the flowers usually contain well developed pistils, and that fruit is subsequently produced.

7. That the Hope vine probably is a staminate vine whose long suppressed pistils have suddenly been regenerated and have recovered the power to function.

8. That the prototype of our present-day rotundifolia vines probably was a true and functioning hermaphrodite.

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PLATE I

Photomicrographs of median sections of flower buds of Vitis rotundifolia

FIG. 1. Scuppernong
FIG. 2. Hope
FIG. 3. Hermaphrodite seedling with reflexed stamens F_1 generation of Scuppernong $\times$ Hope
FIG. 4. Hermaphrodite seedling with upright stamens F_1 generation of Scuppernong $\times$ Hope
FIG. 5. Hermaphrodite seedling with reflexed stamens F_1 generation of Thomas $\times$ Hope
FIG. 6. Hermaphrodite seedling with upright stamens F_1 generation of Thomas $\times$ Hope

PLATE I

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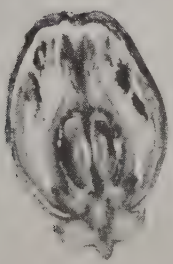

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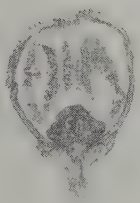
PLATE II

Photomicrographs of median sections of flower buds of Vitis rotundifolia

FIG. 1. Staminate vine (S-10)
FIG. 2. Staminate vine (Light, No. 1)
FIG. 3. Staminate vine (D-21) showing rudiment of a style
FIG. 4-7. Hope, variations in the development of the pistils
FIG. 8. Pistillate rudiment found with some flowers on hermaphrodite vines that bear upright stamens, F_1 generation of Thomas $\times$ Hope
FIG. 9. Pistillate rudiment found with some flowers on hermaphrodite vines that bear the upright stamens, F_1 generation of Scuppernong $\times$ Hope

PLATE II

1



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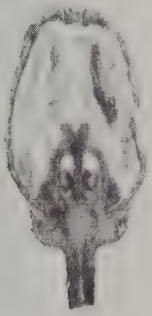

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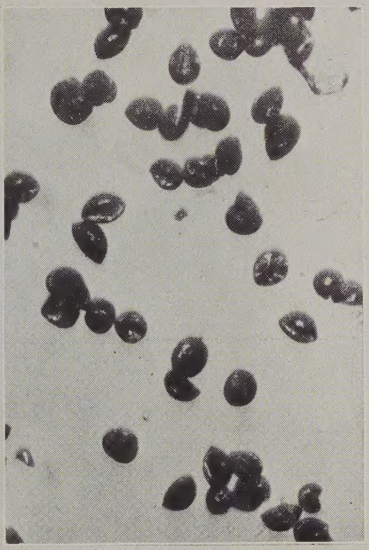
PLATE III

Photomicrographs of pollen grains of Vitis rotundifolia

FIG. 1. Pollen from Hope
FIG. 2. Pollen from Thomas
FIG. 3. Pollen from hermaphrodite seedling with upright stamens, F_1 generations of Thomas × Hope
FIG. 4. Pollen from hermaphrodite seedlings with reflexed stamens, F_1 generation of Thomas × Hope

PLATE III

1



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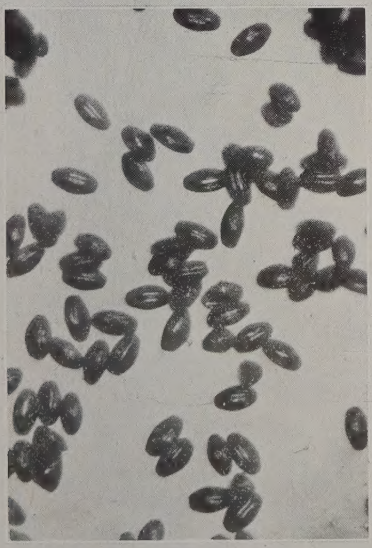

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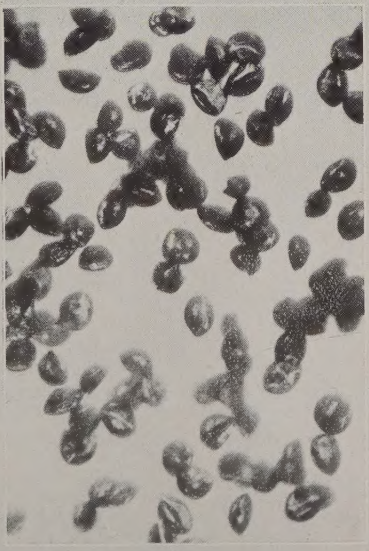

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PLATE IV

Photomicrographs of pollen grains of Vitis rotundifolia

FIG. 1. Pollen from a staminate vine, Dark Male No. 1

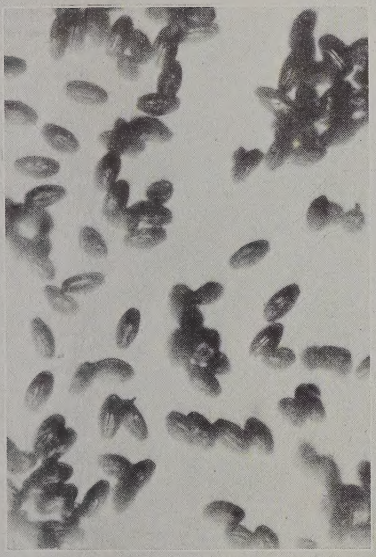
FIG. 2-4. Pollen from the three types of vines of the F_1 generation when Hope is selfed

FIG. 2. Pollen from a staminate vine

FIG. 3. Pollen from a hermaphrodite vine with upright stamens

FIG. 4. Pollen from a hermaphrodite vine with reflexed stamens

PLATE IV



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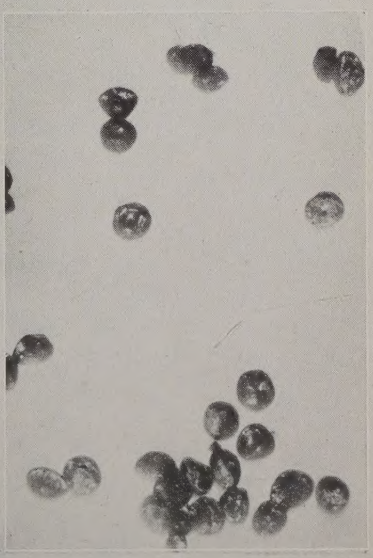

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